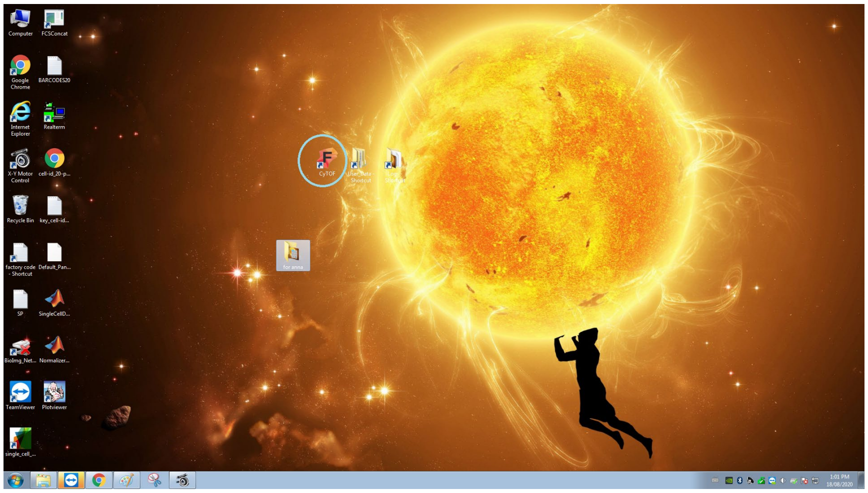
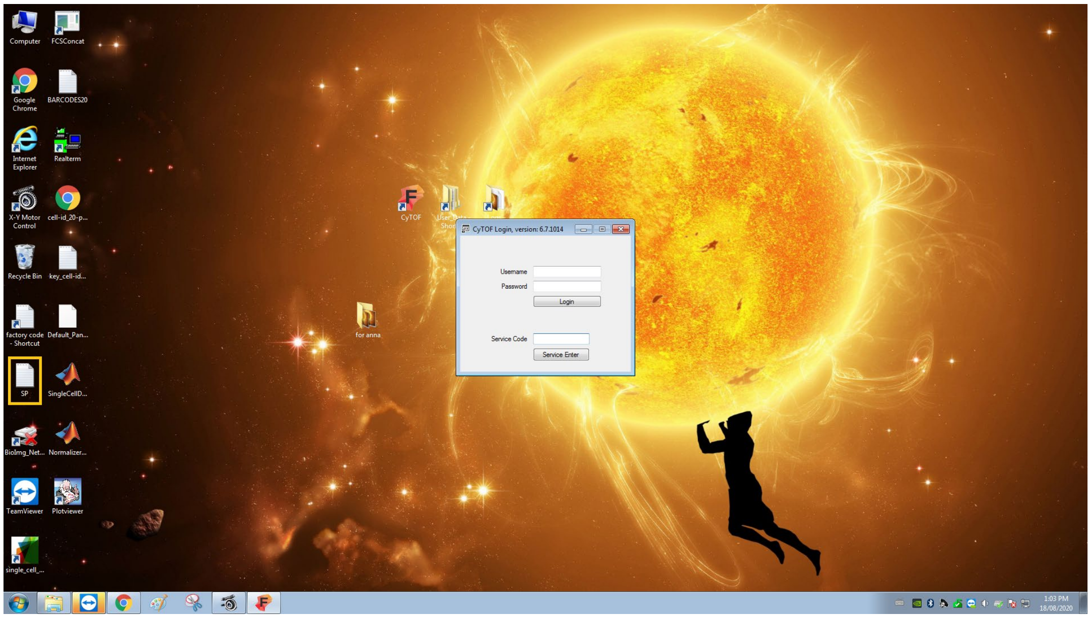
How to turn on the CyTOF?

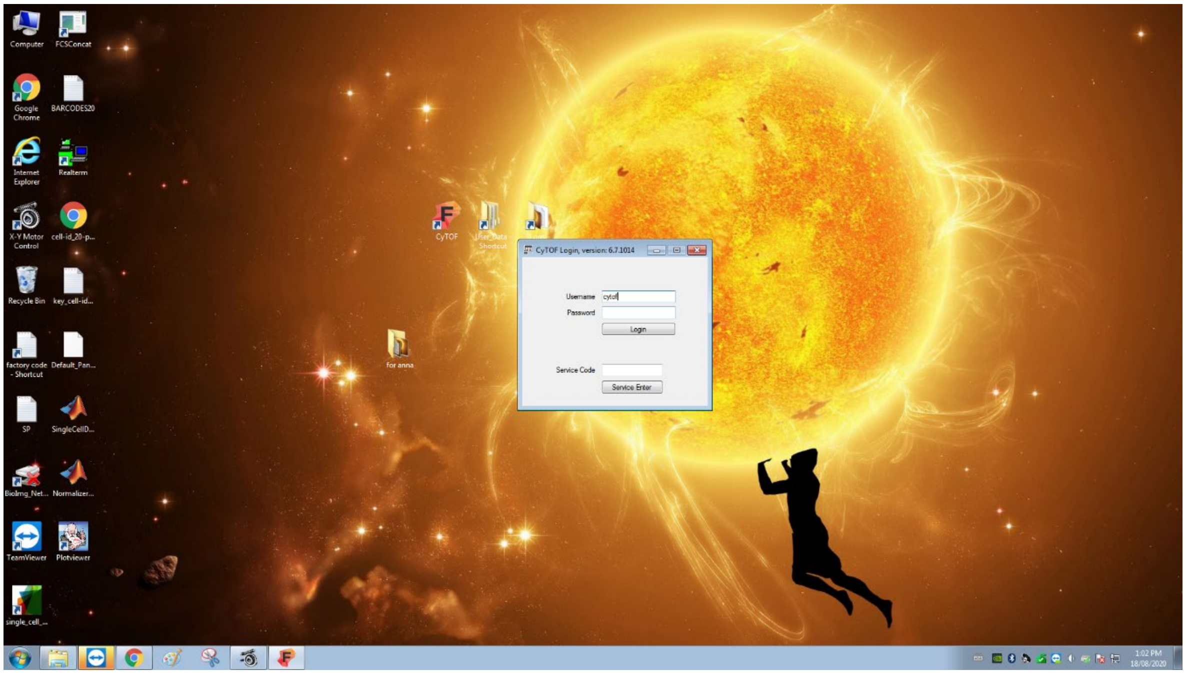
1. Use the username or SP code to get in the program



to get into service mode:



To get into user mode:

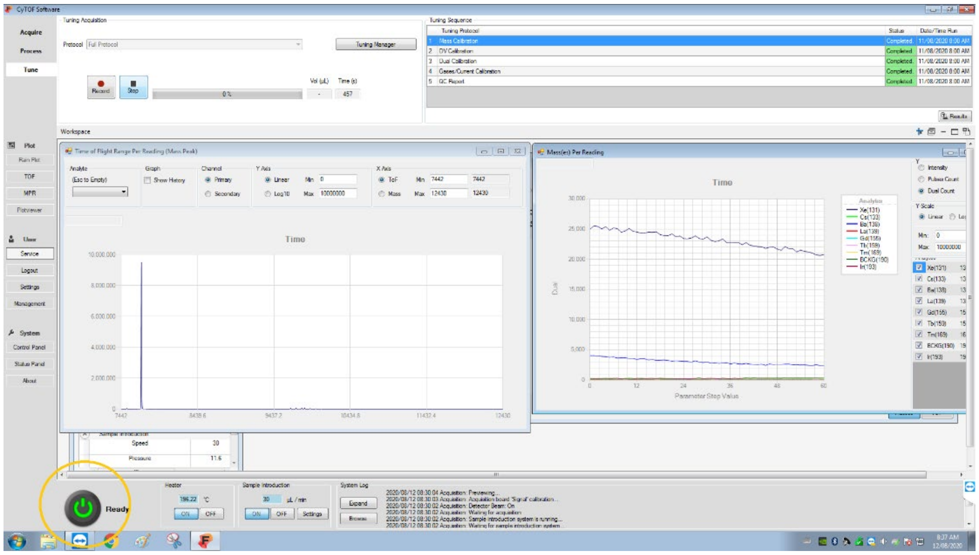


CyTOF Login, version: 6.7.1014

Username: cytof

Password:

2. Turn on the machine (heater and plasma)



Tuning Sequence

	Tuning Protocol	Status	Date/Time Run
1	Mass Calibration	Completed.	11/08/2020 8:00 AM
2	DV Calibration	Completed	11/08/2020 8:00 AM
3	Dual Calibration	Completed.	11/08/2020 8:00 AM
4	Gases/Current Calibration	Completed.	11/08/2020 8:00 AM
5	QC Report	Completed.	11/08/2020 8:00 AM

Heater: 196.22 °C

Sample Introduction: 30 µL / min

Speed: 30

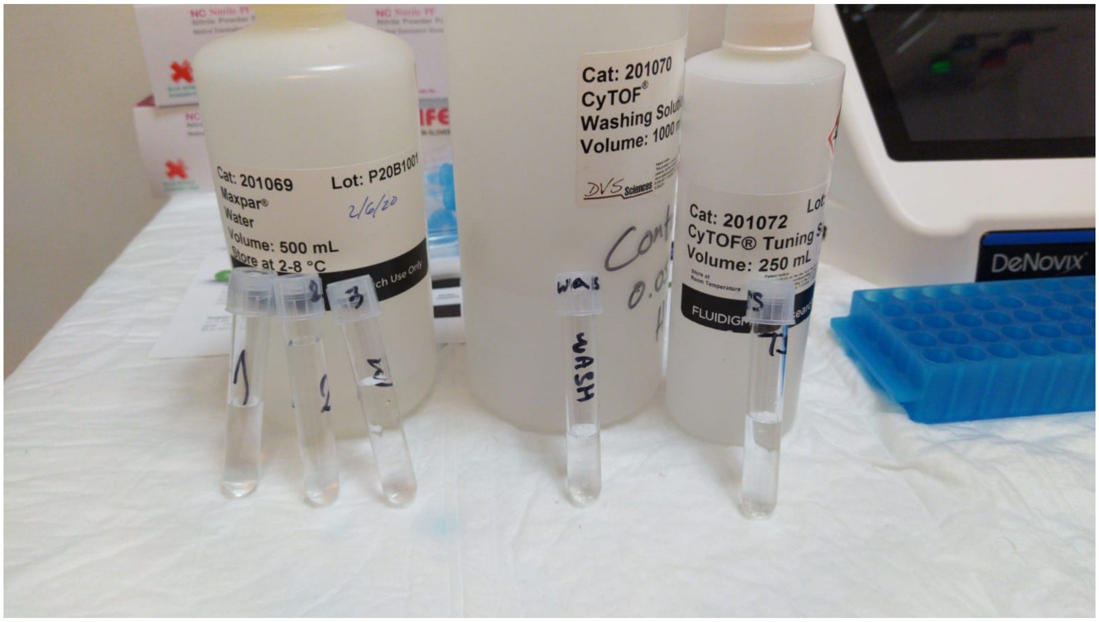
Pressure: 11.6

System Log

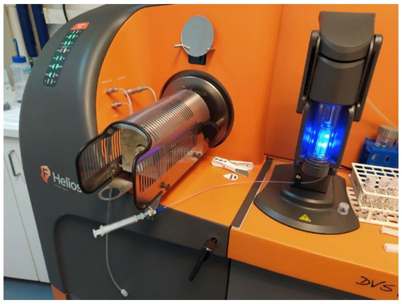
```
2020/08/12 08:30:04 Acquisition: Previewing...
2020/08/12 08:30:03 Acquisition: Acquisition board 'Signal' calibration...
2020/08/12 08:30:02 Acquisition: Detector Beam: On
2020/08/12 08:30:02 Acquisition: Waiting for acquisition
2020/08/12 08:30:02 Acquisition: Sample introduction system is running...
2020/08/12 08:30:02 Acquisition: Waiting for sample introduction system
```

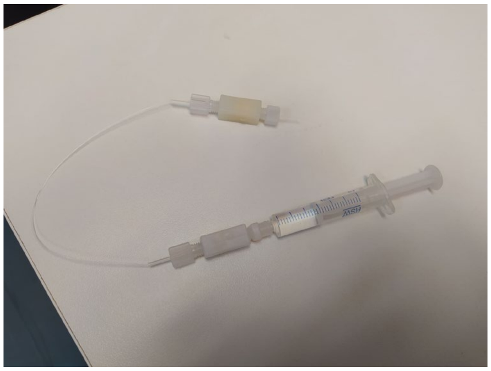
3. Prepare 3 tubes of CyTOF water a tube with tuning solution and another one with washing solution (0.005 HF)

And one 50ml tube with 17.5ml of cytof water (later we add beads).



4.Check the capillaries using the syringes (in and Out)





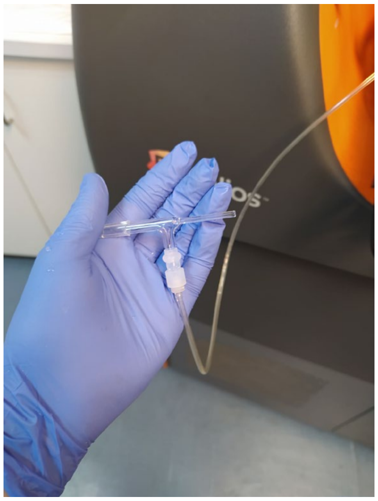
5. Insert water 1 in the machine and press ON for 1 minute. Let it drop on the floor.

6. Place “wash” in the machine to wash the capillaries while you prepare the nebulizer.

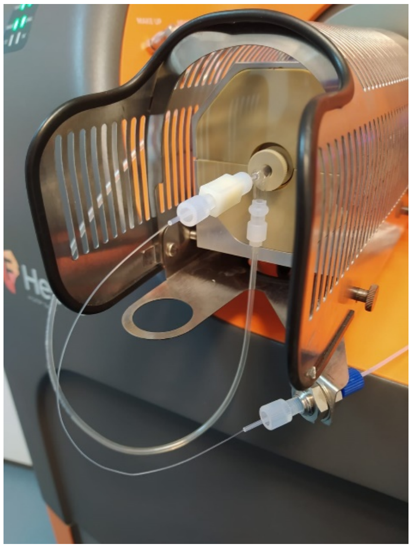
6. Wash the nebulizer

5. Change to “water1” in machine and press ON. And let drop on the floor.

6. Install the nebulizer to the gas pipe and the capillary and turn OFF the plasma.



7. Insert capillary to nebulizer. Turn ON.

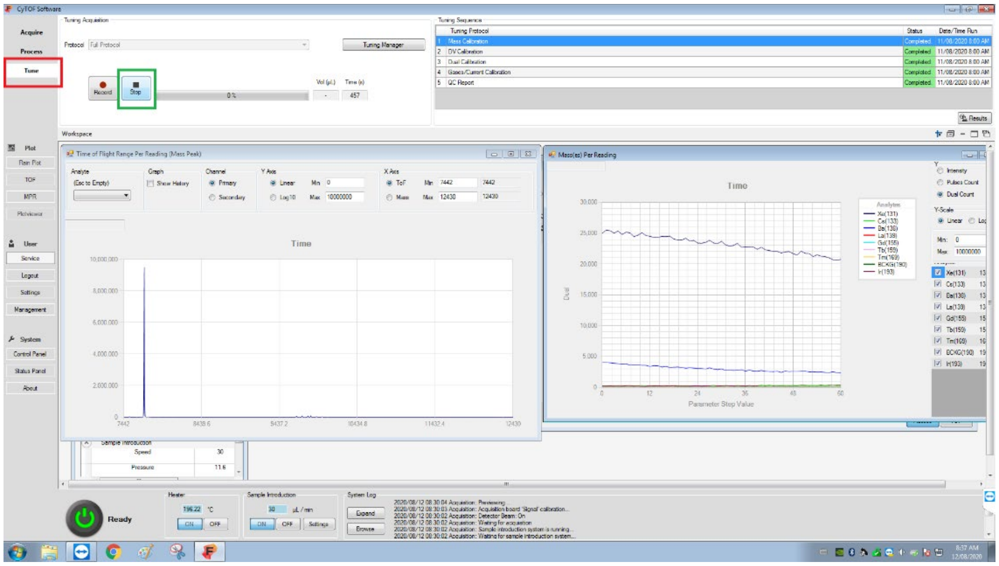


8. Check if the flow is stable on the mirror. If it is, put it inside the CyTOF (add photo)

9. Change to wash for 2-3 minutes.

10. Change back to water1 for 2 minutes.

11. Go to "tune" and preview for water1.



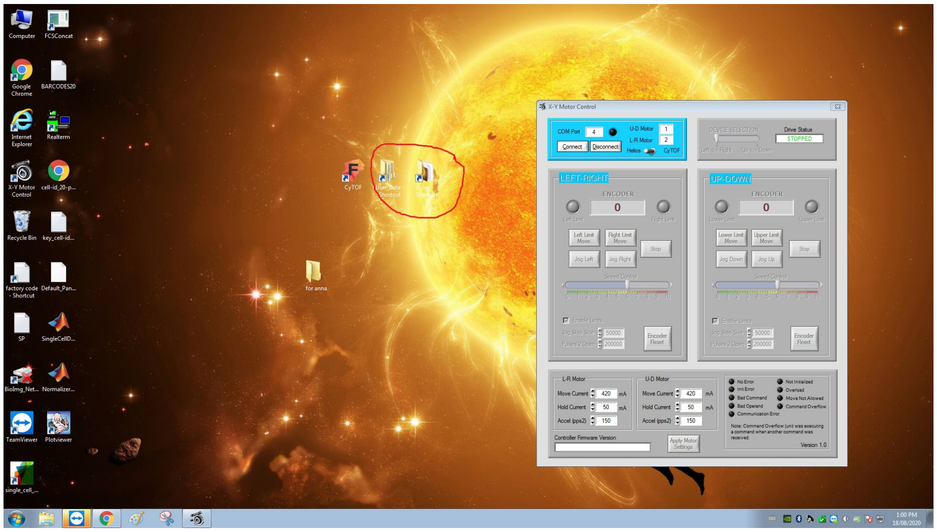
Tuning Sequence

	Tuning Protocol	Status	Date/Time Run
1	Mass Calibration	Completed	11/08/2020 8:00 AM
2	DV Calibration	Completed	11/08/2020 8:00 AM
3	Dual Calibration	Completed	11/08/2020 8:00 AM
4	Gases/Current Calibration	Completed	11/08/2020 8:00 AM
5	QC Report	Completed	11/08/2020 8:00 AM

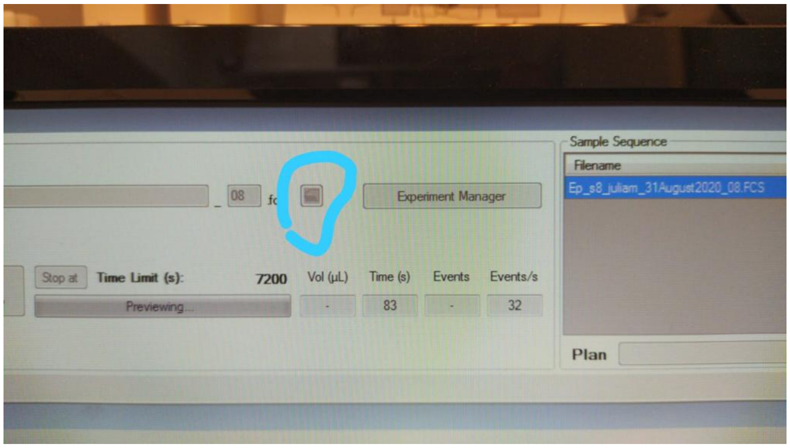
System Log

```
2020/08/12 08:30:04 Acquisition: Previewing...
2020/08/12 08:30:03 Acquisition: Acquisition board 'Signal' calibration...
2020/08/12 08:30:02 Acquisition: Detector Beam: On
2020/08/12 08:30:02 Acquisition: Waiting for acquisition
2020/08/12 08:30:02 Acquisition: Sample introduction system is running...
2020/08/12 08:30:02 Acquisition: Waiting for sample introduction system...
```

12. Prepare the folders of the user. Desktop > User data > Bstomer. Copy the content of New folder (QC,raw etc)> paste in the users folder with today's date (18Augest2020). Create a folder in Logs>082020>18Aug2020



*press and make sure it's in Raw of the user.



Ep_s8_juliam_31August2020_08.FCS

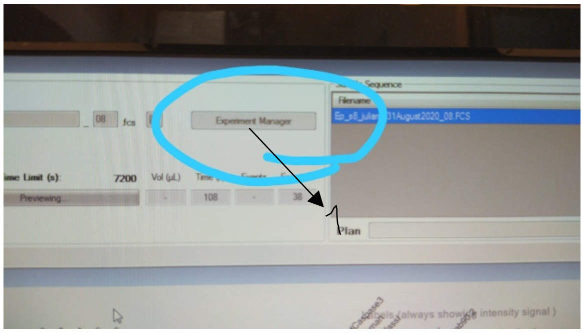
08 fc

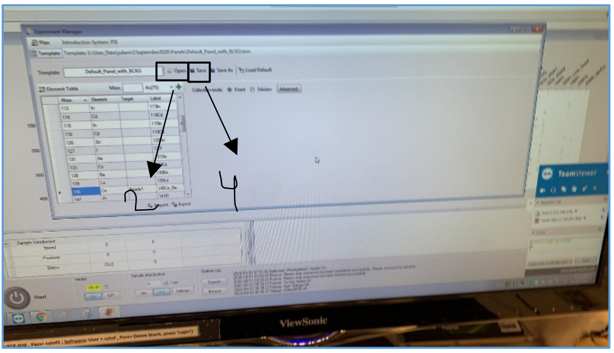
Stop at Time Limit (s): 7200

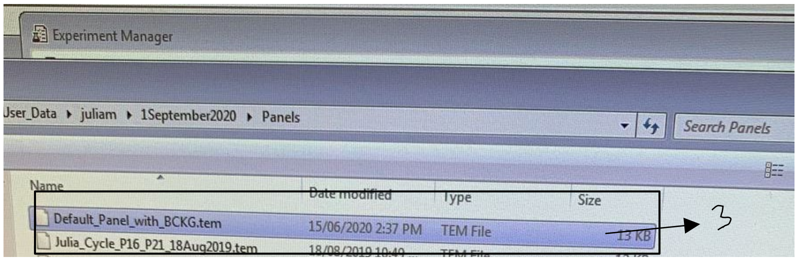
Vol (µL)	Time (s)	Events	Events/s
-	83	-	32

Previewing...

*Press and make sure it's save "Default_Panel_with_BCKG.term" (in Panels of the user)







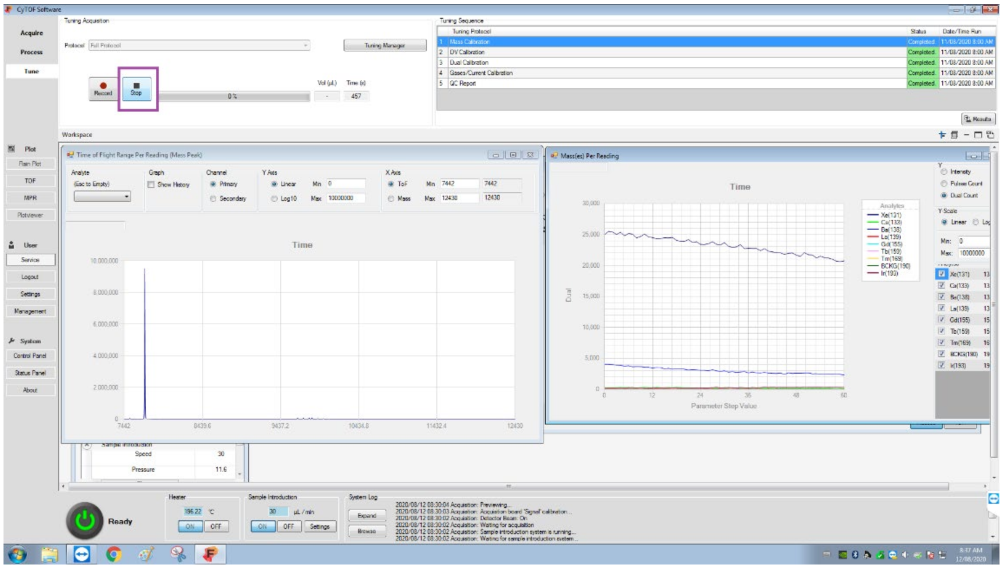
Experiment Manager

User_Data ▸ juliam ▸ 1September2020 ▸ Panels

Search Panels

Name	Date modified	Type	Size
Default_Panel_with_BCKG.tem	15/06/2020 2:37 PM	TEM File	13 KB
Julia_Cycle_P16_P21_18Aug2019.tem	18/08/2019 10:49	TEM File	

13. After the water preview finishes, STOP, put in the Tuning solution (TS) wait for 3 minutes and preview

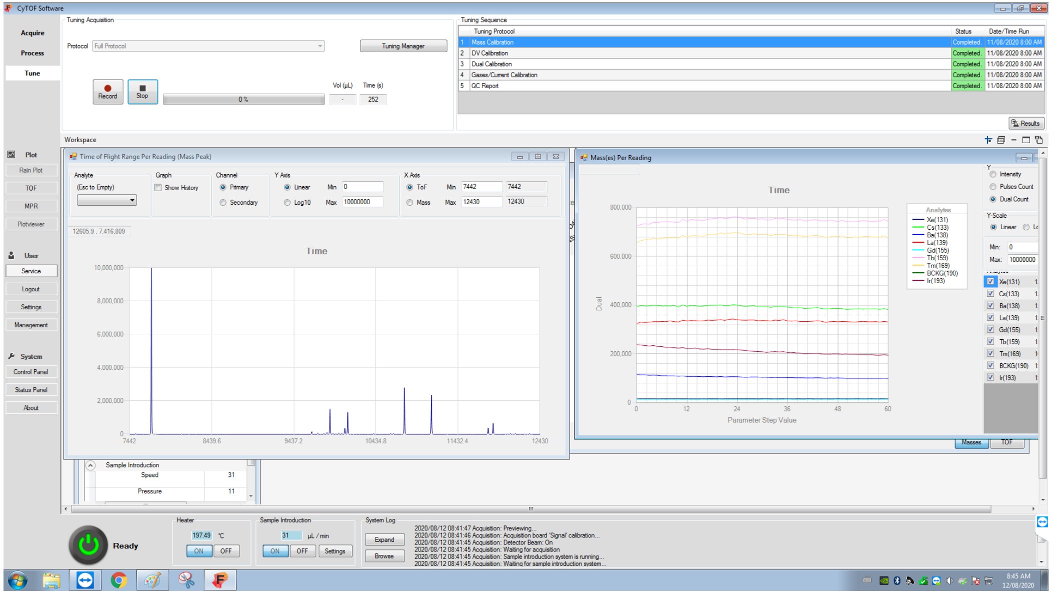


Tuning Sequence

	Tuning Protocol	Status	Date/Time Run
1	Mass Calibration	Completed.	11/08/2020 8:00 AM
2	DV Calibration	Completed.	11/08/2020 8:00 AM
3	Dual Calibration	Completed.	11/08/2020 8:00 AM
4	Gases/Current Calibration	Completed.	11/08/2020 8:00 AM
5	QC Report	Completed.	11/08/2020 8:00 AM

Heater: 196.22 °C
Sample Introduction: 30 µL / min

```
2020/08/12 08:30:04 Acquisition: Previewing...
2020/08/12 08:30:03 Acquisition: Acquisition board 'Signal' calibration...
2020/08/12 08:30:02 Acquisition: Detector Beam: On
2020/08/12 08:30:02 Acquisition: Waiting for acquisition
2020/08/12 08:30:02 Acquisition: Sample introduction system is running...
2020/08/12 08:30:02 Acquisition: Waiting for sample introduction system...
```



Tuning Sequence

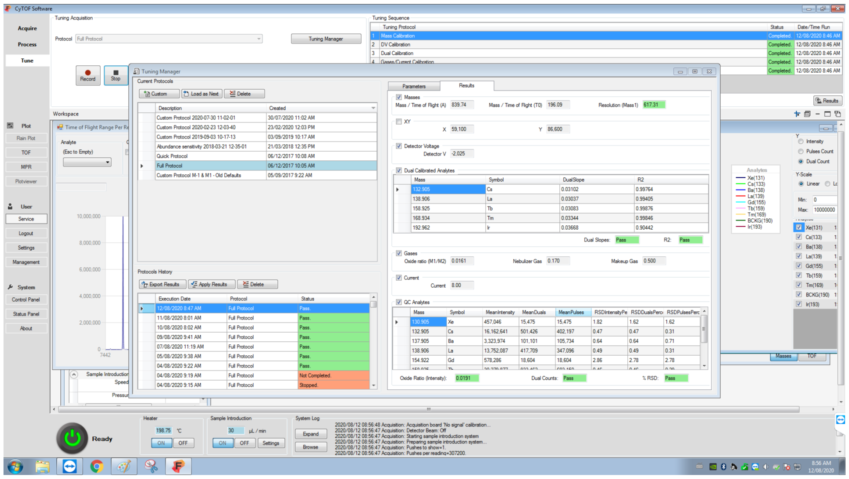
	Tuning Protocol	Status	Date/Time Run
1	Mass Calibration	Completed.	11/08/2020 8:00 AM
2	DV Calibration	Completed.	11/08/2020 8:00 AM
3	Dual Calibration	Completed.	11/08/2020 8:00 AM
4	Gases/Current Calibration	Completed.	11/08/2020 8:00 AM
5	QC Report	Completed.	11/08/2020 8:00 AM

Heater: 197.49 °C
Sample Introduction: 31 µL / min

```
2020/08/12 08:41:47 Acquisition: Previewing...
2020/08/12 08:41:46 Acquisition: Acquisition board 'Signal' calibration...
2020/08/12 08:41:45 Acquisition: Detector Beam: On
2020/08/12 08:41:45 Acquisition: Waiting for acquisition
2020/08/12 08:41:45 Acquisition: Sample introduction system is running...
2020/08/12 08:41:45 Acquisition: Waiting for sample introduction system...
```

14. After the tuning preview is finished press on the record button and wait for the tuning to finish.

15. Snapshot the results



Tuning Manager

Current Protocols

	Description	Created
	Custom Protocol 2020-07-30 11-02-01	30/07/2020 11:02 AM
	Custom Protocol 2020-02-23 12-03-40	23/02/2020 12:03 PM
	Custom Protocol 2019-09-03 10-17-13	03/09/2019 10:17 AM
	Abundance sensitivity 2018-03-21 12-35-01	21/03/2018 12:35 PM
	Quick Protocol	06/12/2017 10:08 AM
▶	Full Protocol	06/12/2017 10:05 AM
	Custom Protocol M-1 & M1 - Old Defaults	05/09/2017 9:22 AM

Protocols History

	Execution Date	Protocol	Status
▶	12/08/2020 8:47 AM	Full Protocol	Pass.
	11/08/2020 8:01 AM	Full Protocol	Pass.
	10/08/2020 8:02 AM	Full Protocol	Pass.
	09/08/2020 9:41 AM	Full Protocol	Pass.
	07/08/2020 11:19 AM	Full Protocol	Pass.
	05/08/2020 9:38 AM	Full Protocol	Pass.
	04/08/2020 9:22 AM	Full Protocol	Pass.
	04/08/2020 9:19 AM	Full Protocol	Not Completed.
	04/08/2020 9:15 AM	Full Protocol	Stopped.

Results

☑ Masses
Mass / Time of Flight (A) 839.74
Mass / Time of Flight (T0) 196.09
Resolution (Mass1) 617.31

☐ XY
X 59,100
Y 86,600

☑ Detector Voltage
Detector V -2,025

☑ Dual Calibrated Analytes

	Mass	Symbol	DualSlope	R2
▶	132.905	Cs	0.03102	0.99764
	138.906	La	0.03037	0.99405
	158.925	Tb	0.03083	0.99876
	168.934	Tm	0.03344	0.99846
	192.962	Ir	0.03668	0.90442

Dual Slopes: Pass
R2: Pass

☑ Gases
Oxide ratio (M1/M2) 0.0161
Nebulizer Gas 0.170
Makeup Gas 0.500

☑ Current
Current 8.00

☑ QC Analytes

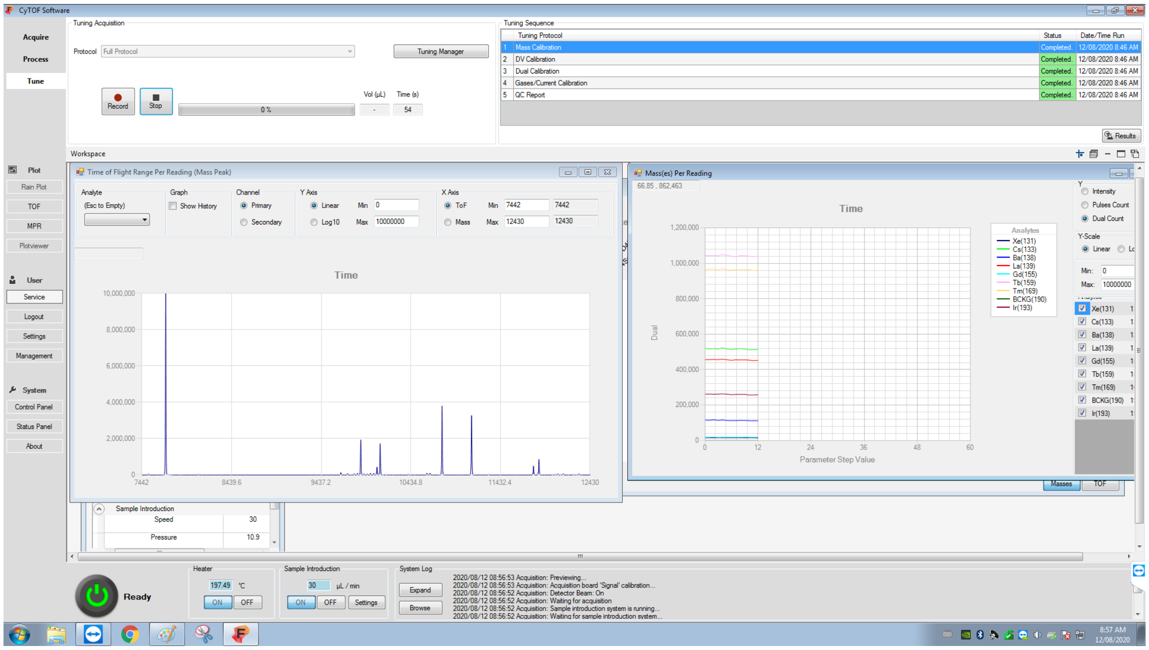
	Mass	Symbol	MeanIntensity	MeanDuals	MeanPulses	RSDIntensityPe	RSDDualsPerc	RSDPulsesPerc
▶	130.905	Xe	457,046	15,475	15,475	1.82	1.62	1.62
	132.905	Cs	16,162,641	501,426	402,197	0.47	0.47	0.31
	137.905	Ba	3,323,974	101,101	105,734	0.64	0.64	0.71
	138.906	La	13,752,087	417,709	347,096	0.49	0.49	0.31
	154.922	Gd	578,286	18,604	18,604	2.86	2.78	2.78

Oxide Ratio (Intensity): 0.0191
Dual Counts: Pass
% RSD: Pass

Heater: 198.75 °C
Sample Introduction: 30 µL / min

```
2020/08/12 08:56:48 Acquisition: Acquisition board 'No signal' calibration...
2020/08/12 08:56:47 Acquisition: Detector Beam: Off
2020/08/12 08:56:47 Acquisition: Starting sample introduction system
2020/08/12 08:56:47 Acquisition: Preparing sample introduction system...
2020/08/12 08:56:47 Acquisition: Pushes to show=1.
2020/08/12 08:56:47 Acquisition: Pushes per reading=307200.
```

16. Preview the tuning solution again for about 2 minutes



Protocol: Full Protocol
Vol (µL): -
Time (s): 54

Analyte: (Esc to Empty)
Graph: ☐ Show History
Channel: ● Primary ○ Secondary
Y Axis: ● Linear ○ Log10 Min 0 Max 10000000
X Axis: ● ToF ○ Mass Min 7442 7442 Max 12430 12430

Y: ○ Intensity ○ Pulses Count ● Dual Count
Y-Scale: ● Linear
Min: 0
Max: 10000000

Tuning Sequence

	Tuning Protocol	Status	Date/Time Run
1	Mass Calibration	Completed.	12/08/2020 8:46 AM
2	DV Calibration	Completed.	12/08/2020 8:46 AM
3	Dual Calibration	Completed.	12/08/2020 8:46 AM
4	Gases/Current Calibration	Completed.	12/08/2020 8:46 AM
5	QC Report	Completed.	12/08/2020 8:46 AM

Sample Introduction
Speed 30
Pressure 10.9

Heater: 197.49 °C ON OFF
Sample Introduction: 30 µL / min ON OFF Settings

System Log

```
2020/08/12 08:56:53 Acquisition: Previewing...
2020/08/12 08:56:53 Acquisition: Acquisition board 'Signal' calibration...
2020/08/12 08:56:52 Acquisition: Detector Beam: On
2020/08/12 08:56:52 Acquisition: Waiting for acquisition
2020/08/12 08:56:52 Acquisition: Sample introduction system is running...
2020/08/12 08:56:52 Acquisition: Waiting for sample introduction system...
```

8:57 AM
12/08/2020

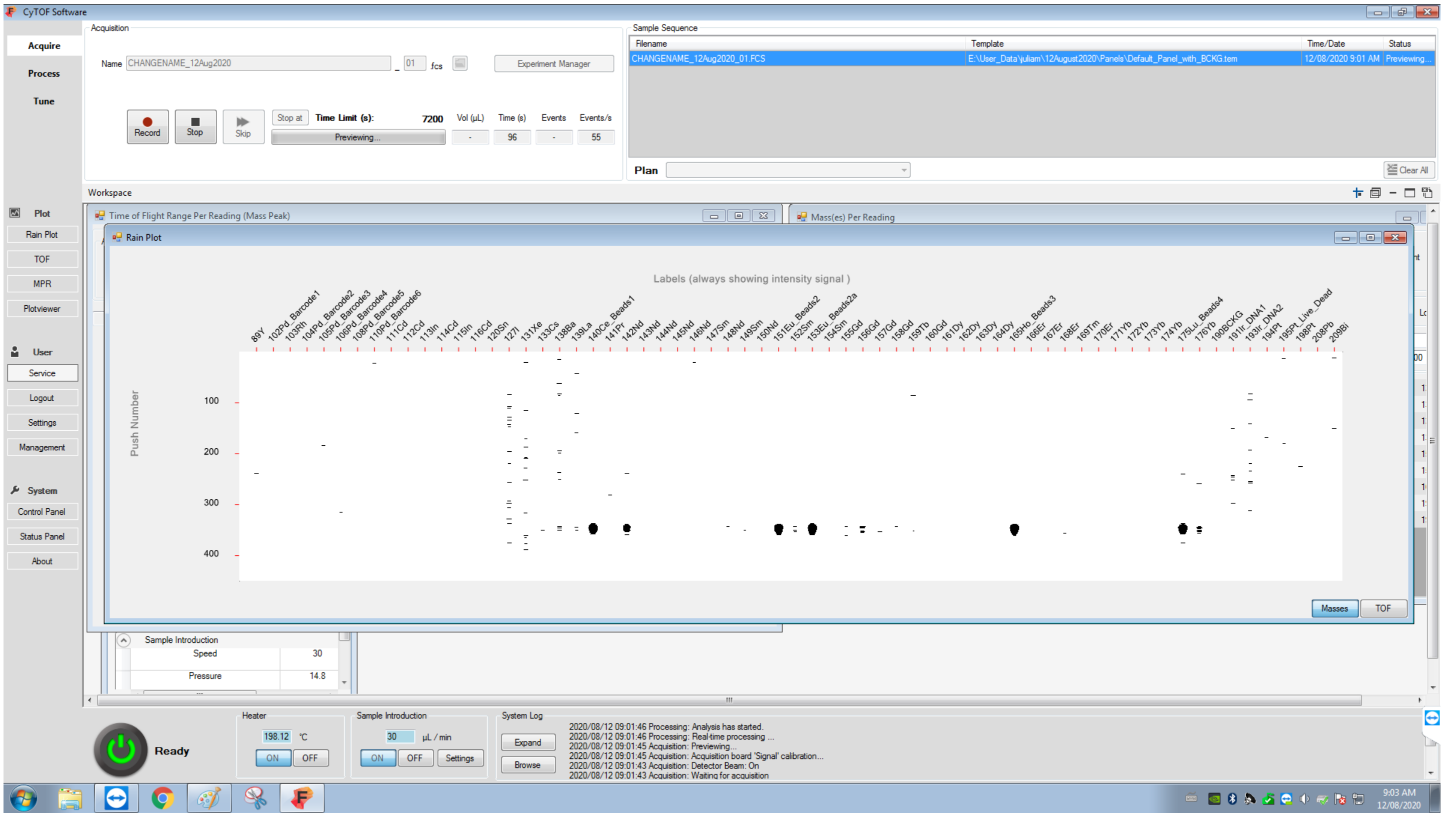
17.Put in ‘water 1’ tube and press on.
18.change for acquire mode and open rainplot.

19. STOP. change to wash. ON. NEVER do preview on wash! Wash for 1 minute.

20. STOP. change back to water1, press ON. Press preview. Make sure events=0. And prepare beads.
21.Take “4 elements calibration beads” from the fridge and vortex them.
22.Put 700ul of beads into a tube with a filter (throw the filter). And add beads to 50ml tube (prepared previously) to sum up to 20ml. *Remembe- the name of the record have to be “BEADS_UserName12Aug2020”

23. Put the tube of filtered beads in the CyTOF.

24. previw and snapshot when events>70 and you see big black dots.



Acquisition

Name CHANGENAME_12Aug2020 _ 01 .fcs

Time Limit (s):	Vol (µL)	Time (s)	Events	Events/s
7200	-	96	-	55

Previewing...

Filename	Template	Time/Date	Status
CHANGENAME_12Aug2020_01.FCS	E:\User_Data\juliam\12August2020\Panels\Default_Panel_with_BCKG.tem	12/08/2020 9:01 AM	Previewing...

Sample Introduction

Speed	30
Pressure	14.8

Heater 198.12 ℃ ON OFF

Sample Introduction 30 µL / min ON OFF Settings

System Log

```
2020/08/12 09:01:46 Processing: Analysis has started.
2020/08/12 09:01:46 Processing: Real-time processing ...
2020/08/12 09:01:45 Acquisition: Previewing...
2020/08/12 09:01:45 Acquisition: Acquisition board 'Signal' calibration...
2020/08/12 09:01:43 Acquisition: Detector Beam: On
2020/08/12 09:01:43 Acquisition: Waiting for acquisition
```

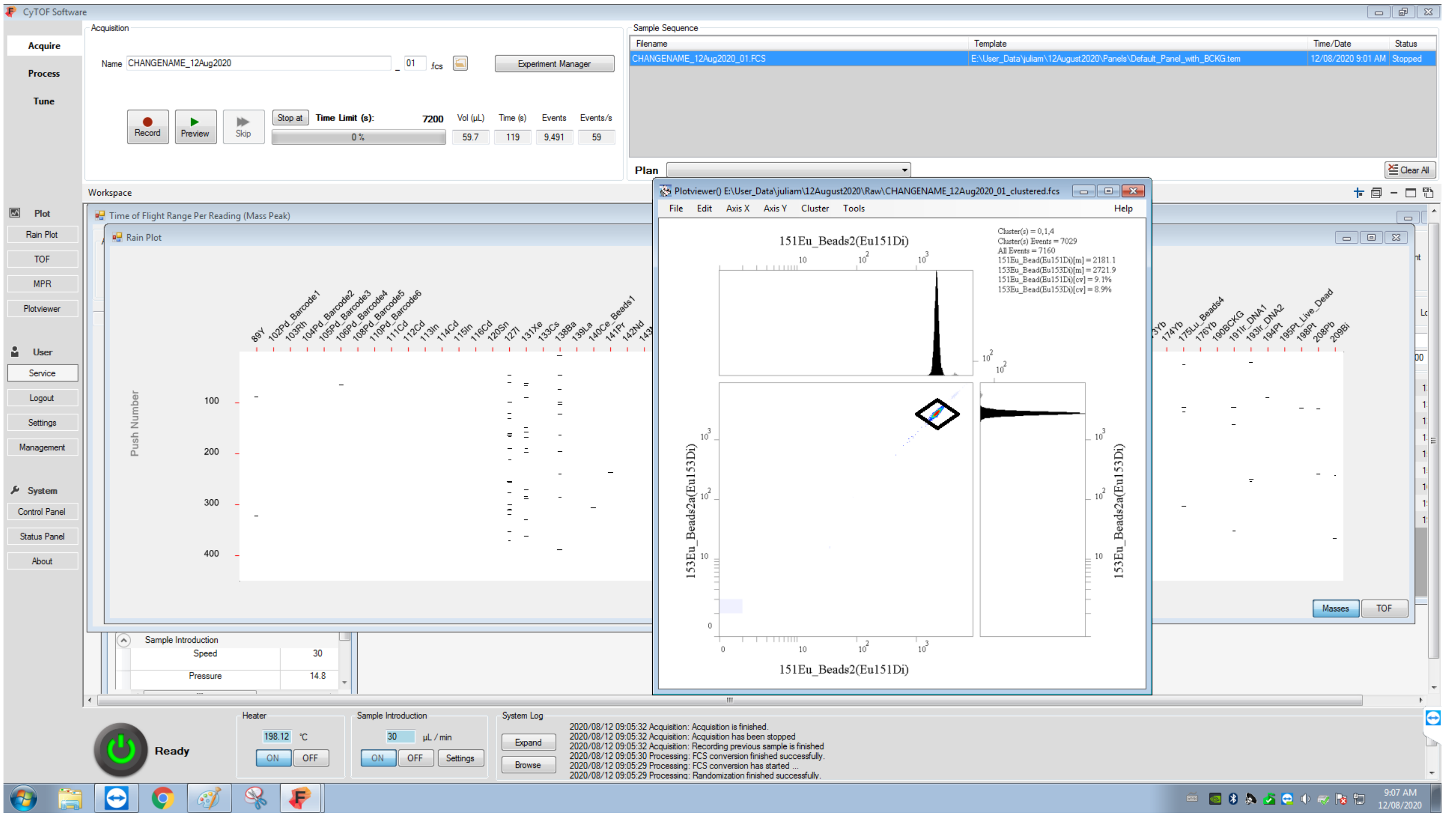
25.press record,record for 120 sec and stop. Change to water2. Press ON.

26. In the plotviewer Open>raw>BEADS12Aug2020.FCS

27.File>cluster current view events

28. Choose 151 and 153 Eu and in X>151. Y>153.

29. Choose only this region (0,1,2,3,4 choose the relevant)



Acquisition

Name CHANGENAME_12Aug2020 _ 01 .fcs

Time Limit (s):	Vol (µL)	Time (s)	Events	Events/s
7200	59.7	119	9,491	59

0 %

Filename	Template	Time/Date	Status
CHANGENAME_12Aug2020_01.FCS	E:\User_Data\juliam\12August2020\Panels\Default_Panel_with_BCKG.tem	12/08/2020 9:01 AM	Stopped

Plotviewer() E:\User_Data\juliam\12August2020\Raw\CHANGENAME_12Aug2020_01_clustered.fcs

```
Cluster(s) = 0,1,4
Cluster(s) Events = 7029
All Events = 7160
151Eu_Bead(Eu151Di)[m] = 2181.1
153Eu_Bead(Eu153Di)[m] = 2721.9
151Eu_Bead(Eu151Di)[cv] = 9.1%
153Eu_Bead(Eu153Di)[cv] = 8.9%
```

Sample Introduction

Speed	30
Pressure	14.8

Heater 198.12 ℃ ON OFF

Sample Introduction 30 µL / min ON OFF Settings

System Log

```
2020/08/12 09:05:32 Acquisition: Acquisition is finished.
2020/08/12 09:05:32 Acquisition: Acquisition has been stopped
2020/08/12 09:05:32 Acquisition: Recording previous sample is finished
2020/08/12 09:05:30 Processing: FCS conversion finished successfully.
2020/08/12 09:05:29 Processing: FCS conversion has started ...
2020/08/12 09:05:29 Processing: Randomization finished successfully.
```

30. Press preview. Make sure events=0.

31. After we see everything looks good, press STOP, and ON (to stop the preview).

32. Change the name from “” to “CHANGENAME_UserName12Aug2020” (fcs= 01)

31. Cut all in folder (the records of tunning and beads) “raw” in user and paste in “QC” (where you saved all snapshots).

32. Copy all in the folder “QC” and past in the folder of today in Logs.